Applicant: Bazan, et al. Attorney's Docket No.: 16622-006001 / DX0903K1

Serial No. : 09/963,347

Filed: September 25, 2001

Page : 2 of 12

AMENDMENTS TO THE SPECIFICATION

Please replace the paragraph on page 57, beginning at line 1, with the following amended paragraph:

Analyses of the expression of cell surface molecules can be performed by direct immunofluorescence. For example, 2 x 10⁵ purified human monocytes are incubated in phosphate buffered saline (PBS) containing 1% human serum on ice for 20 minutes. Cells are pelleted at 200 x g. Cells are resuspended in 20 ml PE or FITC labeled mAb. Following an additional 20 minute incubation on ice, cells are washed in PBS containing 1% human serum followed by two washes in PBS alone. Cells are fixed in PBS containing 1% paraformaldehyde and analyzed on FACScan FACSCAN® flow cytometer (Becton Dickinson; Mountain View, CA). Exemplary mAbs are used, e.g.: CD11b (anti-mac1), CD11c (a gp150/95), CD14 (Leu-M3), CD54 (Leu 54), CD80 (anti-BB1/B7), HLA-DR (L243) from Becton-Dickinson and CD86 (FUN 1; Pharmingen), CD64 (32.2; Medarex), CD40 (mAb89; Schering-Plough France).

Please replace the paragraph on page 57, beginning at line 23, with the following amended paragraph:

For intracytoplasmic staining for cytokines, monocytes are cultured (1 million/ml) in Yssel's medium in the absence or presence of IL-B50 and LPS (E. coli 0127:B8 Difco) and 10 mg/ml Brefeldin A (Epicentre technologies Madison WI) for 12 hrs. Cells are washed in PBS and incubated in 2% formaldehyde/PBS solution for 20 minutes at RT. Subsequently cells are washed, resuspended in permeabilization buffer (0.5% saponin (Sigma) in PBS/BSA (0.5%) /Azide (1 mM)) and incubated for 20 minutes at RT. Cells (2 x 10⁵) are centrifuged and resuspended in 20 ml directly conjugated anti-cytokine mAbs diluted 1:10 in permeabilization buffer for 20 minutes at RT. The following antibodies can be used: IL-1α-PE (364-3B3-14); IL-6-PE (MQ2-13A5); TNFα-PE (MAb11); GM-CSF-PE (BVD2-21C11); and IL-12-PE (C11.5.14; Pharmingen San Diego, CA). Subsequently, cells are washed twice in permeabilization buffer and once in PBS/BSA/Azide and analyzed on FACSean FACSCAN® flow cytometer (Becton Dickinson; Mountain View, CA).

Applicant: Bazan, et al. Attorney's Docket No.: 16622-006001 / DX0903K1

Serial No.: 09/963,347

Filed: September 25, 2001

Page : 3 of 12

Please replace the paragraph on page 62, beginning at line 13, with the following amended paragraph:

and resuspended in an EDTA-containing medium to dissociate the clusters. Viable DC were first counted using trypan blue exclusion of dead cells. Remaining cells were stained with a variety of mouse anti-human FITC-conjugated monoclonal antibodies (mAb) including anti-HLA-DR (Becton Dickinson), anti-CD40, anti-CD80 and CD86 (all from Pharmingen) or an Ig-G1 isotype control (Becton Dickinson), and were analyzed with a FACSean FACSCAN® flow cytometer (Becton Dickinson). Dead cells were excluded based on side and forward scatter characteristics.